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9 IN THE UNITED STATES PATENT AND TRADEMARK OFFICE
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11 TITLE: METHOD FOR PREVENTING FUNGAL
12 GROWTH IN PLANTS
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BACKGROUND OF THE INVENTION

1. Field of The Invention

The present invention relates to horticulture and floriculture, and the prevention or eradication of fungal infestation in particular.

2. Background Information

Fungal infestation is a very significant problem for plant growers. Not only are the fungal species and the plants they attack numerous and varied, the particular chemicals required are also such, and in almost all cases carry some degree of risk to users and all others who come into contact with the chemicals through residual presence, over-spray, and wind drift (in applicable circumstances).

Even though they are currently effective in many instances, even if completely innocuous to humans and animals, the continued viability of existing synthetic chemical fungicides is of concern because of developing resistance of certain fungal species.

Examples of fungal problems to plant growers are limitless, but a few examples will be provided in this context.

A problem facing tomato growers is exemplary of those facing many other food and non-food growers. The recent ban

1 on methyl bromide leaves tomato growers susceptible to, among
2 other problems, fusarium wilts. Some farmers have indicated
3 an intent to simply stop farming, because profitable crops
4 without methyl bromide seems an unlikely prospect.

5 Pythium is a water mold which consumes roots of plants,
6 resulting in stunted growth and eventually plant death.
7 Although phythium is a problem for numerous annuals and
8 perennials, it particularly favors (and destroys) geraniums
9 and poinsettias.

10 Another fungal threat is that from Phytophthora, which
11 causes stem, crown and root rot in summer squash, winter
12 squash (including pumpkins, pepper, tomato, and eggplant), and
13 fruit rots on cucumber, cantaloupe, and watermelon.

14 Still other fungal threats to cash crops include
15 poinsettia scab, English daisy rust, powdery mildew.

16 In each of the cited examples (and most others not so
17 cited), synthetic chemical fungicides are presently the
18 default treatment of choice, simply for lack of a known,
19 viable alternative.

20 Alternatives to conventional synthetic chemical
21 fungicides known as "biofungicides" are increasingly sited as
22 alternatives to the increasingly suspect former class of
23 agents. However, biofungicides are of questionable
24 practicality. Biofungicides are bacteria based, can be very
25 species-specific, and can exhibit serious shelf life

limitations. Disincentives to development and market introduction of biofungicides include EPA registration expenses, which can range from \$300,000 to \$3 million.

In view of the foregoing, it would well serve everyone affected by plant grown (virtually everyone) to provide a wholly new approach to fungal prevention, control, or eradication, one which is innocuous to humans, has no adverse environmental effects, is cost effective, and, of course, is effective.

SUMMARY OF THE INVENTION

It is an object of the present invention to provide a new method for preventing and/or treating fungal infestation of plants.

It is another object of the present invention to provide a new fungicide useful in preventing and/or treating fungal infestation of plants.

It is another object of the present invention to provide a new fungicide useful in preventing and/or treating fungal infestation of plants, which fungicide avoids the dangers associated with presently available synthetic chemical fungicides.

It is another object of the present invention to provide a new fungicide useful in preventing and/or treating fungal infestation of plants, which fungicide is substantially environmentally benign.

1 In satisfaction of these and related objects, the present
2 invention provides a natural fungicide and associated method
3 of use which, as an alternative to conventional use of
4 synthetic chemical fungicides, provides effective prevention
5 and eradication of fungal infestation by a wide spectrum of
6 fungal species.

7 It is believed that the present natural fungicide (a
8 garlic extract) will be exempt from EPA registration rules.
9 This, together with its simple and readily available principle
10 ingredient, renders the present composition a highly cost
11 effective choice in fungal control.

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13 BRIEF DESCRIPTION OF THE DRAWINGS

14 Table 1 depicts laboratory results of investigations
15 designed to test the efficacy of garlic extract treatment of
16 a wide array of fungal species.

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18 DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENT

19 The present invention is of a garlic extract-based,
20 natural fungicide and associated method for preventing and/or
21 treating fungal infestation through the use thereof.

22 To establish the efficacy of the present invention,
23 controlled laboratory evaluations were carried out.

Methodology

The efficacy of the garlic extract was tested against nine (9) soil-borne fungi that cause root and lower stem rots of plants. The fungal pathogens included: *Pythium aphanidermatum*, *Pythium irregular*, *Pythium ultimum*, *Phytophthora capsici*, *Phytophthora cinnamomi*, *Phytophthora nicotiana*, *Rhizoctonia solani*, *Fusarium oxysporum* f.sp. *lycopersici*, *Fusarium oxysporum* f.sp. *spinaciae*, and *Thielaviopsis basicola*. All isolates used in this investigation were known to be pathogenic to various cultivated plant species.

All isolates of *Pythium aphanidermatum*, *Pythium irregular*, *Pythium ultimum*, *Phytophthora capsici*, *Phytophthora cinnamomi*, *Phytophthora nicotiana*, *Rhizoctonia solani*, *Fusarium oxysporum* f.sp. *lycopersici*, *Fusarium oxysporum* f.sp. *spinaciae* were grown on standard corn meal agar (CMA) for 8 to 9 days. Plugs from inoculum plates were taken using a #4 cork borer (5 mm diameter). Inoculum plugs were placed into 5-cm sterile plastic petri dishes containing 10 ml of a basal nutrient solution that consisted of 60% (v/v) clarified V-8 juice and sterile distilled water (SDW). Basal nutrient solutions were amended with the appropriate treatments. Treatments included SDW (non-treated control), a fungicidal

1 agent appropriate to each species (metylaxyl for the Pythium
2 and Phytophthora species, PCNB for Rhizoctonia, and
3 Allegiance(r) for Fusarium), or 10%, 15%, 20%, 25%, or 30%
4 (v/v) garlic extract. Cultures were placed into a growth
5 chamber and maintained in the dark at 22(C(1(C. After 48
6 hours, mycelial growth (if any was visible) from culture plugs
7 was measured.

8 Additionally, plugs were taken from one of the two
9 replicate treatments for each fungal isolate for the
10 non-treated control and the 10% garlic extract treatments,
11 washed in SDW and placed on CMA plates to check for viability
12 of the inoculum after 48 hours exposure to the treatments.
13 Plates were allowed to remain in the growth chamber for eight
14 (8) days. After eight (8) days, any additional growth from the
15 inoculum plug was measured. All treatments were replicated
16 twice and three repetitions of the experiments were performed.

17 For Thielaviopsis basicola, chlamydospores, rather than
18 mycelial growth, were challenged with the treatments.
19 Treatments were as listed above. However, 0.05-ml of a stock
20 solution of chlamydospores were placed in sterile capped
21 centrifuge tubes with 1-ml of the appropriate treatment. The
22 fungicide control for Thielaviopsis basicola was Baytan(r).
23 After 24 hours, tubes were spun down for 10 minutes at 13,000

1 rpm. A 0.1-ml sample of the concentrate was spot plated onto
2 TBCEN agar (selective for *Thielaviopsis basicola* and allows
3 for growth of this obligate parasitic fungus) in duplicate.
4 Concentrates were then washed twice in SDW, being agitated and
5 concentrated as described above. These washed concentrates
6 were plated as described above to determine if the treatment
7 resulted in death of the *Thielaviopsis basicola*
8 chlamydospores. Each treatment was replicated twice.

9 Results and Discussion

10 All *Pythium* and *Phytophthora* species tested grew on the
11 untreated basal nutrient solution (Table 1). However, none of
12 the *Pythium* and *Phytophthora* species placed on basal nutrient
13 solution amended with metalaxyl or garlic extract at 10% to
14 30% grew from the inoculum plugs.

15 The *Rhizoctonia* species tested grew on the untreated
16 basal nutrient solution (Table 1). In the first replication,
17 *Rhizoctonia* grown on the basal nutrient solution amended with
18 PCNB displayed a limited amount of growth due to an
19 inappropriate PCMB concentration. However, in the second
20 replication, *Rhizoctonia* did not grow on the basal nutrient
21 solution amended with PCNB. *Rhizoctonia* placed on the basal
22 nutrient solution amended with garlic extract at 10% to 30%
23 did not grow from the inoculum plugs.

All *Fusarium* species tested grew on the untreated basal nutrient solution (Table 1). However, none of the *Fusarium* species placed on basal nutrient solution amended with Allegiance(r) fungicide or garlic extract at 10% to 30% grew from the inoculum plugs.

For all *Pythium*, *Phytophthora*, *Rhizoctonia* and *Fusarium* species tested, when plugs were removed from the treatment solutions, washed and transferred to CMA, all untreated controls grew from the plugs onto the CMA within 48 hours. However, none of those grown in basal nutrient solution amended with 10% garlic extract grew from the plugs after eight (8) days.

Therefore, garlic extract as low as 10% (v/v) inhibited mycelial growth of all of the fungal pathogens tested in this study. Additionally, even when removed from the presence of the garlic solution, fungal pathogens failed to grow and no sign of viable fungal mycelium could be observed. Thus, the garlic extract was not only fungistatic, but also fungicidal.

Conclusions

The above investigations establish beyond any reasonable doubt that a garlic extract solution is reliably efficacious in preventing and treating fungal infestation. Because the subject composition is wholly devoid of harmful chemical agents, regardless of the context (human or animal health, or

1 general environmental), the present composition and associated
2 method for preventing and treating fungal infestation
3 represents a long-awaited, much-needed alternative to
4 presently available fungicides.

5 Use of the garlic extract in lieu of conventional
6 fungicides is straight forward. One merely sprays a light
7 coating of the garlic extract solution over plants leaves,
8 stems and (optionally) on the nearly soil. This may be
9 repeated daily without any deleterious effects on any known
10 plant, but treatments as infrequently as once weekly have thus
11 far proven effective in for some plant and fungal species.

12 Of course, the means of application are as varied as
13 those for presently known fungicides, and may range from hand-
14 held garden sprayers to fixed plumbing and nozzle assemblies,
15 to aerial application.

16 Further research may establish optimal concentrations of
17 garlic extract for specific plant and fungal species, but
18 present indications are that concentrations as little as 10%
19 (v/v) are adequate for preventing all tested fungal agents.
20 Eradication of existing fungal infestations are thought to
21 require, in some cases, concentrations a bit higher, perhaps
22 in the 20% range.

23 Although the invention has been described with reference
24 to specific embodiments, this description is not meant to be
25 construed in a limited sense. Various modifications of the

1 disclosed embodiments, as well as alternative embodiments of
2 the inventions will become apparent to persons skilled in the
3 art upon the reference to the description of the invention.
4 It is, therefore, contemplated that the appended claims will
5 cover such modifications that fall within the scope of the
6 invention.

Table 1. Growth of fungal species treated with fungicides or varying concentrations of garlic extract.

Fungal pathogen	Treatment^z	Plates with visible mycelial growth (%)	Measured growth (cm)^y
<i>Pythium aphanidermatum</i>			
Fungicide		0	0
Non-treated control		100	2.0
10% garlic		0	0
15% garlic		0	0
20% garlic		0	0
25% garlic		0	0
30% garlic		0	0
Significance		***	***
<i>Pythium irregular</i>			
Fungicide		0	0
No treatment control		100	2.0
10% garlic		0	0
15% garlic		0	0
20% garlic		0	0
25% garlic		0	0
30% garlic		0	0
Significance		***	***
<i>Pythium ultimum</i>			
Fungicide		0	0
No treatment control		100	2.0
10% garlic		0	0
15% garlic		0	0
20% garlic		0	0
25% garlic		0	0
30% garlic		0	0
Significance		***	***
<i>Phytophthora capsici</i>			
Fungicide		0	0
Non-treated control		100	0.6
10% garlic		0	0
15% garlic		0	0
20% garlic		0	0
25% garlic		0	0
30% garlic		0	0
Significance		***	***

Phytophthora cinnamomi

Fungicide	0	0
Non-treated control	100	0.9
10% garlic	0	0
15% garlic	0	0
20% garlic	0	0
25% garlic	0	0
30% garlic	0	0
Significance	***	***

Phytophthora nicotiana

Fungicide	0	0
Non-treated control	100	1.4
10% garlic	0	0
15% garlic	0	0
20% garlic	0	0
25% garlic	0	0
30% garlic	0	0
Significance	***	***

Rhizoctonia solani

Fungicide	50 ^x	0.3
Non-treated control	100	1.7
10% garlic	0	0
15% garlic	0	0
20% garlic	0	0
25% garlic	0	0
30% garlic	0	0
Significance	***	***

Fusarium oxysporum f.sp.
lycopersici

Fungicide	0	0
Non-treated control	100	1.1
10% garlic	0	0
15% garlic	0	0
20% garlic	0	0
25% garlic	0	0
30% garlic	0	0
Significance	***	***

Fusarium oxysporum f.sp.
spinaciae

Fungicide	0	0
Non-treated control	100	1.5
10% garlic	0	0
15% garlic	0	0
20% garlic	0	0
25% garlic	0	0
30% garlic	0	0
Significance	***	***

*** Significant at the P>F level of 0.001.

^x Fungicide controls included Metylaxyl for the *Pythium* and *Phytophthora* species, PCNB for *Rhizoctonia*, Allegiance® for *Fusarium* and Baytan® for *Thielaviopsis*.

^y Growth (cm) from edge of inoculum plug.

^z Limited mycelial growth occurred in the first control replication due to inappropriate concentration of PCNB fungicide.